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Abstract

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Document type : *Article de périodique (Journal article)*

Référence bibliographique

Vicca, Sara ; Zavalloni, Costanza ; Fu , Yongshuo H ; Voets, L ; Dupré de Boulois, Hervé ; et. al. *Arbuscular mycorrhizal fungi may mitigate the influence of a joint rise of temperature and atmospheric CO₂ on soil respiration in Grasslands*. In: *International Journal of Ecology*, Vol. 2009, p. ID 209768 (2009)

DOI : 10.1155/2009/209768

Research Article

Arbuscular Mycorrhizal Fungi May Mitigate the Influence of a Joint Rise of Temperature and Atmospheric CO₂ on Soil Respiration in Grasslands

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Received 24 June 2009; Accepted 16 October 2009

Recommended by Johannes van Veen

We investigated the effects of mycorrhizal colonization and future climate on roots and soil respiration (R_{soil}) in model grassland ecosystems. We exposed artificial grassland communities on pasteurized soil (no living arbuscular mycorrhizal fungi (AMF) present) and on pasteurized soil subsequently inoculated with AMF to ambient conditions and to a combination of elevated CO₂ and temperature (future climate scenario). After one growing season, the inoculated soil revealed a positive climate effect on AMF root colonization and this elicited a significant AMF \times climate scenario interaction on root biomass. Whereas the future climate scenario tended to increase root biomass in the noninoculated soil, the inoculated soil revealed a 30% reduction of root biomass under warming at elevated CO₂ (albeit not significant). This resulted in a diminished response of R_{soil} to simulated climatic change, suggesting that AMF may contribute to an attenuated stimulation of R_{soil} in a warmer, high CO₂ world.

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1. Introduction

In grasslands the majority of the ecosystem carbon pool is stored belowground, making soil respiration (R_{soil}) an important component of the carbon balance of grassland ecosystems [1]. Up to now, R_{soil} remains one of the most uncertain components of carbon cycle models, ranging from process-based ecosystem models [2] to earth system models [3]. Responses of R_{soil} to climate change have been well studied for single factor effects of, for example, warming, CO₂ enrichment, or changes in precipitation [4–6]. However, multifactor manipulative experiments remain scarce and the complexity of interactive effects contributes largely to the limited understanding.

Soil respiration represents the integrated CO₂ flux of root respiration, mycorrhizal respiration (often considered part of the autotrophic respiration), and heterotrophic respiration. Two key factors that control R_{soil} are carbon supply and temperature [4]. Carbon supply depends primarily on plant

productivity, which generally responds positively to CO₂ enrichment [5–7] mainly because of increased photosynthetic rates [8]. Via consequent increases in litterfall [9], root exudation [10], root biomass [11], and root turnover [12], carbon availability for the microbial community increases, stimulating microbial biomass and activity [6]. Moreover, root respiration probably also increases because of the increased root biomass and possibly also because of increased root respiration per unit biomass (specific root respiration) [13]. Overall, CO₂ responses of R_{soil} beneath herbaceous species range between a 10% decline and a 162% increase [14]. Given that all biochemical processes are temperature dependent, warming may increase R_{soil} [15] via enhanced autotrophic [16, 17] and heterotrophic respiration [17]. However, this response is often restricted to the early stage of heating experiments, after which R_{soil} frequently returns to its original level [18]. Such downregulation is likely due to the depletion of labile organic carbon pools in the soil [19–22], reducing microbial activity, but thermal adaptation of

root [23, 24] and microbial activity [19] could also play an important role.

Studies on the effects of a combination of elevated CO₂ and temperature on soil dynamics are scarce and the results of the few studies examining the combined effects of warming and CO₂ enrichment are equivocal [20–22, 25, 26]. Possibly, indirect effects such as changes in soil moisture or nutrient availability are responsible for the variability among the observed responses. Moreover, also the temperature sensitivity (often expressed as Q_{10} , the factor by which R_{soil} is multiplied when temperature increases with 10°C) of R_{soil} may change in response to climatic change. As shown by Wan et al. [26], Q_{10} of R_{soil} might decrease in response to a joint rise of temperature and atmospheric CO₂ concentration, but opposite trends were observed by Tingey et al. [25]. More research is obviously required to solve this issue.

Many manipulation experiments focusing on climate change impacts on ecosystem functioning were conducted under controlled conditions, without considering the mycorrhizal fungi living in symbiosis with the plants. However, in grassland ecosystems, most plants are associated with arbuscular mycorrhizal fungi (AMF) [27]. Hence, an important fraction of R_{soil} may be assigned to AMF. Despite the low decomposability of chitin—the structural backbone of fungal tissue [28]—colonization by AMF can enhance belowground respiration rates [29], although this was not observed in all studies [30]. An AMF-induced increase of R_{soil} could result from respiration of the biotrophs themselves, but indirect AMF effects on root exudation [31], root longevity and decomposition [32], soil aggregate stability [33], and nutrient acquisition [29] are probably crucial as well. In this context, recent research that highlighted the central role of mycorrhizal fungi may play regarding feedbacks on global change [34, 35].

Arbuscular mycorrhizal fungi also are sensitive to climate change. Elevated CO₂ concentrations can indirectly affect AMF through increased C allocation from the host plant to the fungus [36], although this effect might be overestimated under abrupt compared to gradual increases of atmospheric CO₂ [37]. A meta-analysis by Treseder [38] revealed an overall increase in mycorrhizal abundance in response to elevated CO₂. In turn, mycorrhizal fungi can affect plant and soil dynamics. In a review on CO₂ effects on plant symbiosis with mycorrhizal fungi, Diaz [39] reported that most infected plants exhibited an additional increase in dry weight and/or a better nutritional status when exposed to elevated CO₂ concentrations. Furthermore, mycorrhizal hyphae can redistribute the recently fixed carbon away from the roots. Hence, in the presence of mycorrhizal fungi, effects of CO₂ enrichment are not necessarily restricted to the rhizosphere. Moreover, the glycoprotein glomalin that is produced by AMF stimulates soil aggregation [40]. In a high CO₂ world, AMF could thus enhance soil C sequestration directly via C allocation to deeper soil and indirectly via enhanced soil aggregate stability through enhanced glomalin production [41].

Studies combining AMF and climate warming are less abundant. In one study, a temperature-induced increase in fungal growth was associated with increased specific

root length [42]. Hawkes et al. [43] found that plant photosynthates were more rapidly transferred to and respired by AMF when exposed to elevated temperatures. With regard to R_{soil} , however, interactions between warming and/or CO₂ enrichment and the AMF status of plants remain to be tested.

In a model grassland experiment, we exposed plant communities on pasteurized soil (no living AMF present) and on pasteurized soil subsequently inoculated with AMF to ambient conditions and to a joint rise of atmospheric CO₂ and temperature (future climate scenario). We investigated how R_{soil} and root characteristics such as root length, biomass, diameter, and C : N ratio responded to this future climate and to AMF colonization. A major objective was to test for interactions between presence of AMF and the future climate scenario.

2. Materials and Methods

2.1. Study Site and Experimental Set-Up. This study was conducted at the Drie Eiken Campus of the University of Antwerp (Wilrijk, Belgium, 51° 09' N, 04° 24' E). The climate of Northern Belgium is characterized by mild winters and cool summers, with average annual air temperatures (T_{air}) varying around 9.6°C. Annual precipitation averages 776 mm and is more or less equally distributed throughout the year.

In May 2007, an experimental platform with artificially assembled grassland model ecosystems was established. The platform consisted of 10 sunlit chambers, each with an interior surface area of 2.25 m². The top was covered with a colorless polycarbonate plate. The sides were covered with transparent polyethylene film. Five of these chambers were exposed to ambient T_{air} and ambient CO₂ concentrations. The other five chambers were continuously exposed to a future climate scenario with approximately 620 ppm CO₂ and air temperatures were warmed 3°C above fluctuating ambient T_{air} . All future-climate chambers had their individual CO₂ control group. The CO₂ concentration was measured every 8 seconds with an infrared gas analyzer (WMA-4, PP-Systems, Hitchin, UK) and the concentration was adjusted independently in each chamber.

This experiment was performed on 24 grassland communities, spread over the 10 chambers. Hence, each climate scenario x AMF inoculation combination was represented by six replicates. Each community contained 18 plants, with three individuals of six species, selected from three functional groups: two grass species (*Poa pratensis* L., *Lolium perenne* L.), two N-fixing dicots (*Medicago lupulina* L., *Lotus corniculatus* L.), and two non-N-fixing dicots (*Rumex acetosa* L., *Plantago lanceolata* L.). The 18 individuals were planted in a hexagonal grid with a 4.5 cm interspace between the plants. Interspecific interactions were maximized by avoiding clumping.

Each plant community was constructed in a PVC tube with a height of 40 cm and an inner diameter of 24 cm. The PVC tubes were filled to a height of 36 cm with sandy soil (89.2% sand, 8.7% silt, 2.1% clay), collected from an extensively managed grassland in Berlaar (Antwerp, Belgium). The soil was pasteurized at 90°C, during two

successive 8-hour cycles. Subsequently, the soil was well mixed to obtain similar initial microbial communities in all communities. At the moment we transplanted the five-week-old seedlings from seedling trays to the PVC tubes, half of the communities were inoculated with 100 g inoculum, consisting of calcined clay with two AMF taxa, *Gigaspora margarita* MUCL 41833 (+/-2 spores/g of inoculum) and *Glomus intraradices* BEG 34 (40 root fragments per g of inoculum, 85% frequency of root colonization [44]). The inoculum (5.5 g per seedling) was placed in direct contact with the root systems at transplantation. Our experiment thus consisted of four treatments: inoculated pasteurized soil at ambient temperature and CO₂ concentrations (IA), inoculated pasteurized soil at elevated temperature and CO₂ concentrations (IE), non-inoculated pasteurized soil at ambient temperature and CO₂ concentrations (NA), and non-inoculated pasteurized soil at elevated temperature and CO₂ concentrations (NE).

In order to avoid unrealistic soil temperatures, the PVC tubes containing the grassland communities were buried in the soil. During the experiment, the communities in both climate scenarios received equal irrigation amounts. Communities were watered three times per week with a drip irrigation system at a rate of 0.05 L hour⁻¹m⁻². The amount of water supplied was based on the 10-year average monthly precipitation recorded in the nearby meteorological station of Deurne (Antwerp, Belgium). Water could freely drain from the containers, while capillary rise of groundwater was prevented by means of drainage pipes installed beneath the chambers.

2.2. Soil Respiration. In October 2007, five months after the start of the experiment, we measured R_{soil} on all 24 plant communities of each treatment. Measurements were made on five days between 15 and 22 October. During each of these five days, we measured R_{soil} in each community three times within 10 minutes, using an infrared gas analyzer (EGM-4; PP-Systems, Hitchin, UK) coupled to a small PVC chamber (8 × 5 × 1.5 cm). These PVC chambers were permanently installed on a strip of bare soil inside the communities. An aerating hole in the chambers ensured mixing with the outside air to avoid build-up of CO₂ concentrations inside the chambers and was closed with terostat (Henkel KGaA, Düsseldorf, Germany) during the measurements of R_{soil} . In order to avoid confounding effects through changes in plant photosynthesis [45], R_{soil} was always measured in the morning (between 8 hours and 12 hours, with measurements of the different treatments randomized).

One of our objectives was to compare the temperature sensitivities of R_{soil} in the different treatments. To this end, we altered the temperatures in the chambers ca. 24 hours before measuring R_{soil} , such that we obtained a sufficiently large soil temperature range as well as sufficient overlap in the temperature ranges between both climate scenarios. In the ambient chambers, R_{soil} was measured at soil temperatures (at 5 cm depth) of, on average, 7.7, 9.2, 10.9, 12.2, and 14°C, while average soil temperatures under the future climate scenario were 7.7, 9.1, 11.1, 13.5, and 14.8°C during R_{soil} measurements.

2.3. Soil and Root Sampling. Six months after the start of the experiment, belowground biomass was harvested. Twelve soil cores (2.0 cm diameter) were taken in four of the six plant communities of each treatment. The entire soil profile of each community was sampled and divided into four depths: 0–9, 10–18, 19–27, and 28–36 cm. In order to obtain comparable soil samples, soil cores were collected following the same pattern for each community: we sampled six cores near each of the six species present and six other cores were sampled in between three species, considering different species combinations. All 12 samples from the same depth in the same community were pooled and a subsample was used for root analyses. We washed the extracted root material by hand and stored the roots in a Ringer's solution in the fridge (4°C). Within two weeks after sampling, roots were analyzed for root length and diameter, using WinRHIZO image analysis software (Regent Instruments Inc., Quebec, Canada). Subsequently, roots were oven dried at 70°C for 48 hours before being weighed. We determined the C and N concentrations on 5 to 7 mg roots from the top 9 cm layer, using a C/N analyzer (NC-2100, CarloErba, Italy). Further, we also analyzed 25 to 35 mg soil of each soil layer (0–9 cm, 10–18 cm, 19–27 cm, and 28–36 cm; dried at 60°C for 48 hours) of each community for C and N concentration.

2.4. Mycorrhizal Root Colonization. In order to quantify the degree of AMF root colonization, dried root samples (from the top 9 cm soil of four plant communities per treatment) were cleared in a 10% KOH solution (90°C for 30 minutes) and then stained with a blue ink solution (90°C for 30 minutes; 1% HCl with 2% blue ink, Parker; adapted from Vierheilig et al. [46]). Thirty randomly selected root pieces (10 mm length) of each sample were examined under a bright-field microscope at 50x or 125x magnification. The frequency and intensity of AMF root colonization were estimated according to the method described by Declerck et al. [47]. We calculated the frequency of AMF colonization as the percentage of root segments that contained hyphae, arbuscules, or vesicles. The intensity of colonization, that is, the abundance of vesicles, hyphae, and arbuscules in each colonized root segment, was estimated using different intensity classes (1%–20%, 21%–40%, 41%–60%, 61%–80%, 81%–100%) [47].

2.5. Potential Heterotrophic Respiration. Air-dried soil (50 g from the top 9 cm) of 18 plant communities (five samples for IA and NA, four samples for IE and NE) was rewetted to obtain a water content of 60% of field capacity and was subsequently incubated in plastic bottles (1.2 L) at 20°C. We measured respiration rates (potential heterotrophic respiration; R_{hpot}) after 6, 11, 13, and 19 days of incubation, using an infrared gas analyzer (EGM-4; PP-Systems, Hitchin, UK), coupled to the bottles. Each sample was measured twice within five minutes and the average of both measurements was used for further analysis.

2.6. Data Analysis. In order to detect differences in R_{soil} between our treatments, we needed to determine soil respiration at a reference temperature (i.e., the basal soil respiration;

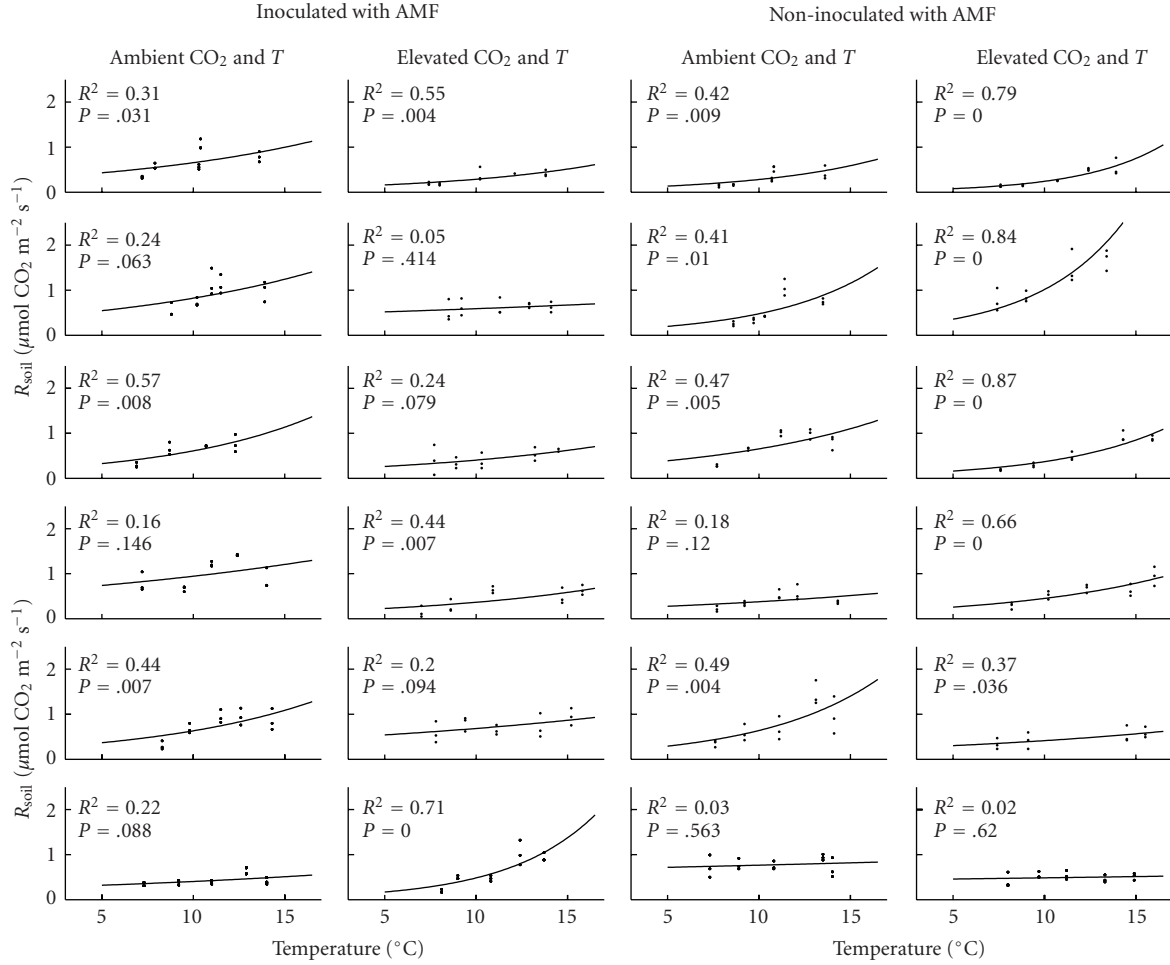


FIGURE 1: Soil respiration (R_{soil}) in function of soil temperature, for all replicates of each treatment. Dots are measured values; the line represents the regression fitted with (1).

BR). As shown by, for example, Wan et al. [26], temperature sensitivity may also change in response to climatic change. We computed the basal respiration rate and the temperature sensitivity (Q_{10}) of R_{soil} , by fitting the following equation to the data:

$$R_{\text{soil}} = \text{BR} \times Q_{10}^{((T-b)/10)} \quad (1)$$

with T being soil temperature at 5 cm depth and $b = 10^\circ\text{C}$ for the ambient treatment or $b = 13^\circ\text{C}$ for the future climate scenario. Like this, we obtained BR at growth conditions (i.e., with a 3°C difference between both climate scenarios). To illustrate the goodness of fit, we present the fitted regressions in Figure 1. For each treatment, we computed the weighted mean BR at growth temperature and the weighted mean Q_{10} , using the inverse of the standard error (SE) on the estimated parameters as weight factors (i.e., $1/(\text{SE of BR})$ and $1/(\text{SE of } Q_{10})$ to compute the weighted mean BR and Q_{10} , resp.).

In this study, with a combination of elevated temperature and CO_2 , we cannot distinguish between thermal acclimation and acclimation to CO_2 enrichment (in single factor experiments, acclimation to, e.g., warming is tested for by

comparing ambient and heated treatments at one common temperature). In our case, only the BR at growth conditions may give an impression of potential downregulation (i.e., lower R_{soil} under the future climate scenario than expected from measurements at ambient conditions), with homeostasis occurring if this BR is equal for both climate scenarios. Homeostasis would thus imply that climate scenario has no effect on R_{soil} . Therefore, we will only discuss differences in BR at growth conditions, that is, BR at 10°C for the ambient treatment versus BR at 13°C for the future climate scenario. The same accounts for the potential heterotrophic respiration. Therefore, we computed R_{hpot} at 23°C for the future climate scenario (for all treatments, R_{hpot} was measured at 20°C), to obtain R_{hpot} with a 3°C difference between both climate scenarios. To this end, we used a Q_{10} of two and computed R_{hpot} at 23°C for the simulated future climate scenario using (1) with $b = 23^\circ\text{C}$.

Regression analyses were performed in Matlab (7.2.0.232, The Mathworks, Natick, MA, USA); statistical analyses were made in SAS (SAS system 9.1, SAS Institute, Cary, NC, USA). Except when stated differently, we used a nested two-way ANOVA to test for effects of AMF inoculation (AMF effects),

TABLE 1: Root and soil characteristics for the different treatments (mean and one standard error (SE)). Letters indicate significant differences within one row ($P \leq .10$; nested two-way ANOVA). Absence of letters denotes the absence of significant differences for the specific parameter. We also report (borderline) significant treatment (AMF or climate scenario) effects and AMF x climate scenario interactions.

		Inoculated pasteurized soil				Non-inoculated pasteurized soil				Statistic results for AMF x climate scenario interaction effects
		Ambient climate		Future climate		Ambient climate		Future climate		
		Mean	SE	Mean	SE	Mean	SE	Mean	SE	
Root biomass (mg cm ⁻³)	Top 9 cm	4.14	1.28	1.64	0.22	3.43	1.04	4.46	1.09	$F_{1,4} = 3.39$; $P = .14$
	Entire profile	1.78 ^{ab}	0.47	1.22 ^a	0.12	1.63 ^{ab}	0.33	2.71 ^b	0.43	$F_{1,4} = 5.72$; $P = .08$
Root length (cm cm ⁻³)	Top 9 cm	35.24	4.33	20.98	3.27	34.91	7.46	36.62	3.39	No
	Entire profile	17.99	3.04	16.64	2.06	18.63	3.33	20.37	2.78	No
Specific root length (cm mg ⁻¹)*	Top 9 cm	10.28	2.06	12.70	0.26	9.87	1.69	9.58	2.01	No
	Entire profile	11.65	0.88	13.12	0.84	10.59	1.10	7.96	1.17	$F_{1,3} = 3.98$; $P = .14$
Average root diameter (mm)	Top 9 cm	0.24	0.01	0.24	0.02	0.21	0.01	0.24	0.01	No
	Entire profile	0.22	0.01	0.23	0.01	0.22	0.00	0.23	0.00	No
Root C concentration (%)	Top 9 cm	42.61	0.93	42.44	0.51	43.37	0.47	42.57	0.66	No
Root N concentration (%)	Top 9 cm	0.93	0.04	1.05	0.05	1.08	0.05	0.99	0.06	$F_{1,4} = 4.37$; $P = .10$
Root C : N ratio	Top 9 cm	46.17	0.90	40.84	1.56	40.33	1.81	43.19	1.78	$F_{1,4} = 6.92$; $P = .06$
Soil C concentration (%)	Top 9 cm	1.32	0.05	1.33	0.08	1.32	0.06	1.33	0.10	No
	Entire profile	1.30	0.04	1.33	0.02	1.29	0.02	1.32	0.03	No
Soil N concentration (%)	Top 9 cm	0.12	0.01	0.12	0.00	0.12	0.00	0.12	0.01	No
	Entire profile	0.12	0.00	0.12	0.00	0.12	0.00	0.12	0.00	No
Soil C : N ratio	Top 9 cm	11.30	0.15	11.23	0.52	11.00	0.23	11.07	0.29	No
	Entire profile	11.09	0.14	11.17	0.32	11.12	0.13	11.06	0.11	No

*Specific root length was calculated from root biomass and root length.

climate effects and AMF x climate scenario interactions, with chamber nested within climate scenario. Differences are reported significant at $P \leq .10$. When not mentioned, the interaction was not significant. In order to test whether AMF root colonization frequency and intensity in the inoculated pasteurized soil significantly differed between both climate scenarios, data were arcsin $\sqrt{(x/100)}$ transformed before performing a one-way ANOVA.

3. Results

3.1. AMF Root Colonization. In our experiment, both pasteurization and inoculation were successful. Throughout the season, only one replicate of the non-inoculated pasteurized soil showed minor AMF colonization (Zavalloni, unpublished). At the end of the season (November 2007), inoculation had resulted in 10% (SE = 3.6) and 20% (SE = 5.8) root colonization frequency and intensity, respectively, in the top 9 cm soil of the ambient treatment. Under warming and CO₂ enrichment, AMF root colonization frequency and intensity in the top 9 cm soil were on average 27.5% (SE = 6.3) and 30.7% (SE = 6.0), respectively. Differences in AMF root colonization frequency differed significantly between both climate scenarios ($F_{1,6} = 5.75$; $P = .05$; one-way ANOVA), whereas colonization intensity did not significantly differ between both treatments ($F_{1,6} = 1.69$; $P = .24$).

3.2. Root and Soil Characteristics. The entire profile root biomass revealed a significant AMF x climate scenario interaction, and a similar tendency was observed for root biomass and length of the top 9 cm soil (Table 1). Hence, the climate effect on root biomass differed between inoculated and non-inoculated pasteurized soil. Whereas we observed a tendency towards a negative climate effect in the inoculated pasteurized soil, root biomass, and root length in the non-inoculated pasteurized soil were higher under the future climate scenario (albeit not significantly).

Despite similar average root diameters across the treatments (Table 1), specific root length (SRL) showed a borderline significant AMF x climate scenario interaction effect (Table 1). Whereas SRL was slightly higher in IE than in NE (for the entire profile), SRL showed no difference between both soils at ambient conditions.

This pattern of AMF determining the climate response recurred in the root C : N ratio. Although neither root C or N concentrations nor root C : N ratio responded significantly to inoculation or to warming and CO₂ enrichment (Table 1), we found a borderline significant AMF x climate scenario interaction for root N concentration and a significant AMF x climate scenario interaction for root C : N ratio (Table 1). Whereas the inoculated pasteurized soil exhibited a tendency towards a positive climate effect on root N concentration, we observed a slight decrease in NE as compared to NA.

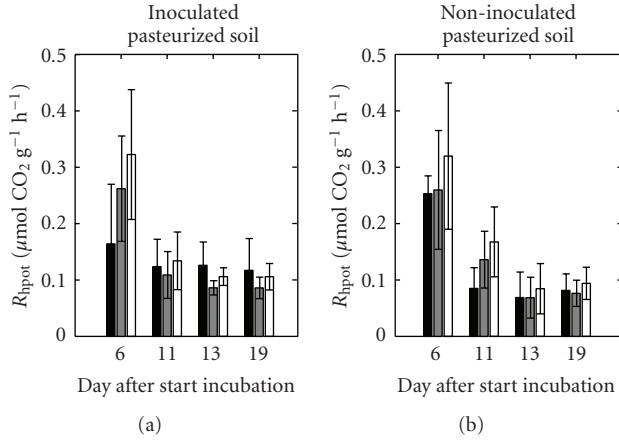


FIGURE 2: Mean potential heterotrophic respiration (R_{hpot}) measured at 20°C for the inoculated and non-inoculated pasteurized soil under ambient conditions (black bars) and under combined warming and CO₂ enrichment (grey bars). For the future climate scenario, we also computed R_{hpot} at 23°C (white bars), using (1) with $b = 23^\circ\text{C}$ and $Q_{10} = 2$, such that we could compare R_{hpot} at growth conditions (for more details, see Section 2). Error bars represent the standard deviation on the mean.

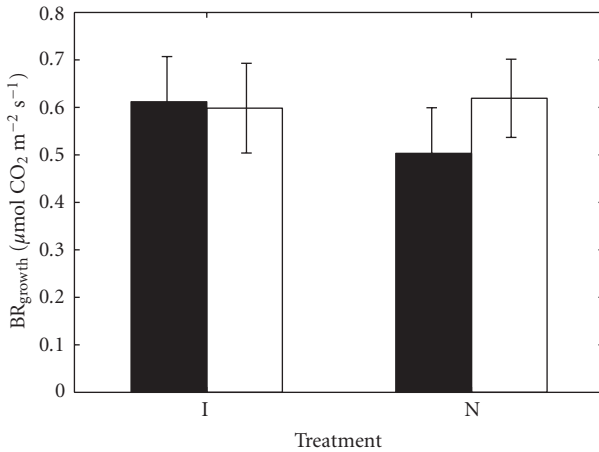


FIGURE 3: Weighted mean basal soil respiration (weighted using the inverse of the standard error of BR) at growth conditions (BR_{growth}) in inoculated pasteurized soil (I) and non-inoculated pasteurized soil (N). Black bars represent BR at 10°C for the ambient climate scenario, and white bars represent BR at 13°C for the future climate scenario (for more details, see Section 2). Error bars are the standard deviation on the weighted mean.

The opposite pattern was, logically, observed for root C : N ratio. Soil C and N concentrations were similar across all treatments (Table 1).

3.3. Potential Heterotrophic Respiration. For all treatments, R_{hpot} decreased over time (Figure 2), reflecting the expected decrease of labile carbon during the incubation period. After six days of incubation, R_{hpot} at growth conditions (black and white bars in Figure 2) was higher under the future climate scenario than under ambient conditions (significant scenario

TABLE 2: Weighted mean Q_{10} (weighted using $1/(\text{SE of } Q_{10})$) and one standard error (SE) on WM for the different treatments. The Q_{10} was calculated by fitting (1) to the data.

	Ambient climate		Future climate	
	Q_{10}	SE	Q_{10}	SE
Inoculated pasteurized soil	2.10	0.13	2.12	0.13
Non-inoculated pasteurized soil	2.05	0.13	2.57	0.12

effect: $F_{1,7} = 5.69$; $P = .05$). Differences between inoculated and non-inoculated pasteurized soil were not statistically significant.

3.4. Soil Respiration. In the inoculated pasteurized soil, basal soil respiration at growth conditions was similar in IA and IE (Figure 3), indicating a homeostasis of R_{soil} . The non-inoculated pasteurized soil revealed a trend towards an increased basal rate at growth conditions under the future climate scenario as compared to ambient conditions (Figure 3), but effects of climate scenario and AMF inoculation were not significant (weighted two-way ANOVA). The Q_{10} of R_{soil} varied around two in all treatments (Table 2); a weighted two-way ANOVA revealed no significant climate scenario or AMF effects.

4. Discussion

4.1. Root Characteristics. In our study, a joint rise in temperature and atmospheric CO₂ did not significantly alter root length or biomass. However, the significant AMF x climate scenario interaction effect implies that the climate effect differed between non-inoculated and inoculated pasteurized soil. The tendency towards a positive climate effect on root biomass in the non-inoculated pasteurized soil agrees with a previous study on the combined effect of warming and CO₂ enrichment [48] (but see [49]) and is probably primarily due to the elevated CO₂ concentrations. Whereas CO₂ enrichment usually enhances root growth and biomass [11, 50, 51], warming either decreases [52–54] or does not affect root biomass [49, 51, 55]. According to Fitter et al. [24], root growth is determined more by resource availability and source-sink relationships than by dominant environmental parameters such as temperature. This may clarify why in the non-inoculated soil root biomass tended to be higher at the future climate scenario than at ambient conditions, a response frequently ascribed to a CO₂-induced increase in carbon supply and belowground allocation, but usually not found under elevated temperatures.

In contrast to the non-inoculated pasteurized soil, root length and biomass in the inoculated pasteurized soil tended to be lower under elevated CO₂ and warming than under ambient conditions. We believe that this significantly different climate effect between inoculated and non-inoculated soil is related to the positive climate effect on AMF root colonization. It is well known that AMF act as a sink for carbon; in exchange for nutrients, carbon is transferred from

the roots to the fungal symbiont. Furthermore, colonization by mycorrhizal fungi frequently resulted in reduced root biomass [56–58], although not all species showed such response [58, 59]. Hence, in the inoculated pasteurized soil, the extra carbon supply at the future climate scenario as compared to ambient conditions was likely transferred to the AMF, in turn reducing root biomass. This postulation is supported by measurements of photosynthesis and aboveground biomass. Whereas the future climate scenario stimulated photosynthetic rates, aboveground biomass was similar across all treatments (Zavalloni, unpublished). Hence, the extra carbon gained under warming and CO₂ enrichment was not invested in aboveground biomass and was most likely transferred to AMF. In the non-inoculated pasteurized soil, carbon could not be transferred to a symbiotic partner, and plants could not rely on AMF for their nutrient supply. Consequently, plants in NE invested more carbon in their roots than plants in IE, which resulted in a significantly higher root biomass in NE as compared to IE.

4.2. Soil Respiration. Our study revealed no significant changes of R_{soil} in response to warming and CO₂ enrichment. The inoculated pasteurized soil even revealed a homeostasis of R_{soil} , as BR at growth conditions was nearly equal for both climate scenarios. This contrasts with most CO₂ enrichment studies that revealed an increased R_{soil} in response to elevated CO₂ [14]. Warming also was frequently observed to enhance R_{soil} [15], although thermal acclimation is often occurs (e.g., [60]). Responses of R_{soil} to a joint rise in temperature and atmospheric CO₂ have been rarely investigated (and most studies were performed on trees), and results of the few studies available are inconsistent [20, 22, 25, 26]. Wan et al. [26] demonstrated that factors such as soil moisture may be responsible for this variability. In our study, soil water content at the end of the growing season did not differ across the treatments (Zavalloni, unpublished). Hence, the observed homeostasis of R_{soil} was unlikely due to water limitations.

The three components of R_{soil} , that is, root, mycorrhizal, and heterotrophic respiration may all alter in response to environmental changes and it is the combination of all three components that caused the homeostasis of R_{soil} . Given the positive climate effect on AMF root colonization, mycorrhizal respiration was likely higher in IE than in IA. Also heterotrophic respiration was probably higher in IE than in IA, as R_{hpot} responded positively to the simulated future climate scenario. Root respiration, the last component of R_{soil} , is determined by root biomass and by specific root respiration (i.e., respiration per unit of root biomass). Specific root respiration is often positively related to root N concentration. Because root N concentration did not significantly differ between IE and IA, we assume that the specific root respiration was similar for both treatments. Hence, decreased root respiration as a consequence of lower root biomass in IE than in IA (–30%, but statistically not significant) is the only component of R_{soil} that can clarify the homeostasis observed in the inoculation pasteurized soil.

Similar to the inoculated soil, the non-inoculated pasteurized soil also did not exhibit a significant difference in

R_{soil} between both climate scenarios, suggesting a potential homeostasis. In contrast to the inoculated pasteurized soil, however, a homeostasis of R_{soil} was unlikely in the non-inoculated pasteurized soil, as BR at growth conditions tended to be circa 20% higher in NE than in NA. Possibly, R_{soil} was too heterogeneous to detect a significant difference between NE and NA with six replicates. In conclusion, we found no real indication for homeostasis of R_{soil} in the non-inoculated pasteurized soil, in particular because both root biomass and R_{hpot} tended to increase in NE as compared to NA. This supports our statement that the positive climate effect on AMF root colonization in the inoculated pasteurized soil induced the homeostasis of R_{soil} via a reduction in root biomass, as neither a homeostasis of R_{soil} , nor an AMF-induced reduction in root biomass was apparent in the non-inoculated pasteurized soil.

Both inoculated and non-inoculated pasteurized soil exhibited similar soil respiration rates. Apparently, AMF did not stimulate R_{h} , as R_{hpot} was similar for AMF treatments. Furthermore, in the simulated future climate, root biomass was significantly lower in IE than in NE. This decrease was, however, not reflected in R_{soil} , which was similar for inoculated and non-inoculated pasteurized soil. Presumably, mycorrhizal respiration, which occurred only in IE, not in NE, and perhaps also specific root respiration (root N concentration was slightly higher in IE than in NE), compensated for the lower root biomass and associated root respiration in IE as compared to NE. Likewise, Cavagnaro et al. [30] concluded that effects of an AMF-induced decrease in root biomass and root length density on R_{soil} were compensated by higher respiration rates of mycorrhizal roots per unit weight as compared to non-mycorrhizal roots.

Last, the temperature sensitivity of R_{soil} was similar across all treatments. The lack of a climate effect on Q_{10} is in agreement with Wan et al. [26], who observed no change in the Q_{10} of R_{soil} under warming and/or CO₂ enrichment in wet conditions, but under dry conditions they observed a tendency towards a positive warming effect on Q_{10} of R_{soil} at both ambient and elevated CO₂ [26]. The similar Q_{10} in IE and IA gives further support to our statement that the homeostasis of R_{soil} was primarily due to the lower root biomass in IE, as differences in substrate quality or substrate limitation would have been reflected in the Q_{10} [61]. Further, we found no AMF effect on the Q_{10} of R_{soil} , supporting earlier observations from Baath and Wallander [62] and Langley et al. [29], who found no effect of AMF root colonization on the temperature sensitivity of root-derived respiration or soil respiration.

5. Conclusions

This study assessed the role of AMF in belowground carbon cycling under current and possible future climate conditions. Combined warming and CO₂ enrichment led to increased AMF root colonization. AMF inoculation and the simulated future climate conditions interacted significantly in their effects on root biomass of artificial six-species grassland communities. Reduction of root biomass upon AMF inoculation resulted in a declined soil respiration

response to a joint rise of temperature and atmospheric CO₂. This may suggest that AMF can contribute to an attenuated stimulation of R_{soil} under a scenario of both rising atmospheric CO₂ and temperature.

Acknowledgments

This project was supported by the Belgian Science Policy-Program 'Science for a Sustainable Development' under contract MYCARBIO, no. SD/BD/05A. The authors thank Joke Van den Berge, Birgit Gielen, and especially Fred Kockelbergh for their valuable help during the set-up of the experiment. They thank Piergiorgio Stevanato for his help with the root analyses, Nadine Calluy and Kristine Crous for the C and N analyses, and Stefan Van Dongen for statistical help. Sara Vicca holds a grant from the Institute for Promotion of Innovation through Science and Technology in Flanders (IWT-Vlaanderen). Costanza Zavalloni is a beneficiary of a Marie Curie International Reintegration Grant (contract MIRG-CT-2005-031109), which partially financed this study. H. Dupré de Boulois is a beneficiary of a grant of Chargé de recherches du F.R.S – FNRS (Belgium). They also thank two anonymous reviewers and the subject editor for providing constructive comments on a previous version of the article. Last, I wish to thank Lola for being my lucky angel.

References

- [1] M. Bahn, M. Rodeghiero, M. Anderson-Dunn, et al., "Soil respiration in European grasslands in relation to climate and assimilate supply," *Ecosystems*, vol. 11, no. 8, pp. 1352–1367, 2008.
- [2] H. Verbeeck, R. Samson, A. Granier, P. Montpied, and R. Lemeur, "Multi-year model analysis of GPP in a temperate beech forest in France," *Ecological Modelling*, vol. 210, no. 1–2, pp. 85–103, 2008.
- [3] P. Friedlingstein, P. Cox, R. Betts, et al., "Climate-carbon cycle feedback analysis: results from the (CMIP)-M-4 model intercomparison," *Journal of Climate*, vol. 19, no. 14, pp. 3337–3353, 2006.
- [4] J. W. Raich and A. Tufekcioglu, "Vegetation and soil respiration: correlations and controls," *Biogeochemistry*, vol. 48, no. 1, pp. 71–90, 2000.
- [5] E. Casella, J. F. Soussana, and P. Loiseau, "Long-term effects of CO₂ enrichment and temperature increase on a temperate grass sward—I: productivity and water use," *Plant and Soil*, vol. 182, no. 1, pp. 83–99, 1996.
- [6] J. A. Morgan, D. R. Lécain, A. R. Mosier, and D. G. Milchunas, "Elevated CO₂ enhances water relations and productivity and effects gas exchange in C3 and C4 grasses of the Colorado shortgrass steppe," *Global Change Biology*, vol. 7, no. 4, pp. 451–466, 2001.
- [7] H. W. Polley, H. B. Johnson, and J. D. Derner, "Increasing CO₂ from subambient to superambient concentrations alters species composition and increases above-ground biomass in a C₃/C₄ grassland," *New Phytologist*, vol. 160, no. 2, pp. 319–327, 2003.
- [8] M. Stitt, "Rising CO₂ levels and their potential significance for carbon flow in photosynthetic cells," *Plant Cell and Environment*, vol. 14, pp. 741–762, 1991.
- [9] P. A. Niklaus, M. Wohlfender, R. Siegwolf, and C. Körner, "Effects of six years atmospheric CO₂ enrichment on plant, soil, and soil microbial C of a calcareous grassland," *Plant and Soil*, vol. 233, no. 2, pp. 189–202, 2001.
- [10] E. Pendall, A. R. Mosier, and J. A. Morgan, "Rhizodeposition stimulated by elevated CO₂ in a semiarid grassland," *New Phytologist*, vol. 162, no. 2, pp. 447–458, 2004.
- [11] H. H. Rogers, G. B. Runion, and S. V. Krupa, "Plant responses to atmospheric CO₂ enrichment with emphasis on roots and the rhizosphere," *Environmental Pollution*, vol. 83, no. 1–2, pp. 155–189, 1994.
- [12] K. S. Pregitzer, D. R. Zak, W. M. Loya, N. J. Karberg, J. S. King, and A. J. Burton, "The contribution of root-rhizosphere interactions to biogeochemical cycles in a changing world," in *The Rhizosphere: An Ecological Perspective*, Z. G. Cardon and J. L. Whitbeck, Eds., pp. 155–178, Academic Press, San Diego, Calif, USA, 2007.
- [13] N. T. Edwards and R. J. Norby, "Below-ground respiratory responses of sugar maple and red maple saplings to atmospheric CO₂ enrichment and elevated air temperature," *Plant and Soil*, vol. 206, no. 1, pp. 85–97, 1999.
- [14] D. R. Zak, K. S. Pregitzer, J. S. King, and W. E. Holmes, "Elevated atmospheric CO₂, fine roots and the response of soil microorganisms: a review and hypothesis," *New Phytologist*, vol. 147, no. 1, pp. 201–222, 2000.
- [15] L. E. Rustad, J. L. Campbell, G. M. Marion, et al., "A meta-analysis of the response of soil respiration, net nitrogen mineralization, and aboveground plant growth to experimental ecosystem warming," *Oecologia*, vol. 126, no. 4, pp. 543–562, 2001.
- [16] A. J. Burton and K. S. Pregitzer, "Field measurements of root respiration indicate little to no seasonal temperature acclimation for sugar maple and red pine," *Tree Physiology*, vol. 23, no. 4, pp. 273–280, 2003.
- [17] X. Zhou, S. Wan, and Y. Luo, "Source components and interannual variability of soil CO₂ efflux under experimental warming and clipping in a grassland ecosystem," *Global Change Biology*, vol. 13, no. 4, pp. 761–775, 2007.
- [18] R. Hyvonen, G. I. Agren, S. Linder, et al., "The likely impact of elevated [CO₂], nitrogen deposition, increased temperature and management on carbon sequestration in temperate and boreal forest ecosystems: a literature review," *New Phytologist*, vol. 173, no. 3, pp. 463–480, 2007.
- [19] M. A. Bradford, C. A. Davies, S. D. Frey, et al., "Thermal adaptation of soil microbial respiration to elevated temperature," *Ecology Letters*, vol. 11, no. 12, pp. 1316–1327, 2008.
- [20] D. Comstedt, B. Bostrom, J. D. Marshall, et al., "Effects of elevated atmospheric carbon dioxide and temperature on soil respiration in a boreal forest using $\delta^{13}\text{C}$ as a labeling tool," *Ecosystems*, vol. 9, no. 8, pp. 1266–1277, 2006.
- [21] G. Lin, P. T. Rygielwicz, J. R. Ehleringer, M. G. Johnson, and D. T. Tingey, "Time-dependent responses of soil CO₂ efflux components to elevated atmospheric [CO₂] and temperature in experimental forest mesocosms," *Plant and Soil*, vol. 229, no. 2, pp. 259–270, 2001.
- [22] B. Pajari, "Soil respiration in a poor upland site of Scots pine stand subjected to elevated temperatures and atmospheric carbon concentration," *Plant and Soil*, vol. 168–169, no. 1, pp. 563–570, 1995.
- [23] D. R. Bryla, T. J. Bouma, and D. M. Eissenstat, "Root respiration in citrus acclimates to temperature and slows during drought," *Plant, Cell and Environment*, vol. 20, no. 11, pp. 1411–1420, 1997.

- [24] A. H. Fitter, J. D. Graves, G. K. Self, T. K. Brown, D. S. Bogie, and K. Taylor, "Root production, turnover and respiration under two grassland types along an altitudinal gradient: influence of temperature and solar radiation," *Oecologia*, vol. 114, no. 1, pp. 20–30, 1998.
- [25] D. T. Tingey, E. H. Lee, R. Waschmann, M. G. Johnson, and P. T. Rygielwicz, "Does soil CO₂ efflux acclimatize to elevated temperature and CO₂ during long-term treatment of Douglas-fir seedlings?" *New Phytologist*, vol. 170, no. 1, pp. 107–118, 2006.
- [26] S. Wan, R. J. Norby, J. Ledford, and J. F. Weltzin, "Responses of soil respiration to elevated CO₂, air warming, and changing soil water availability in a model old-field grassland," *Global Change Biology*, vol. 13, no. 11, pp. 2411–2424, 2007.
- [27] D. J. Read and J. Perez-Moreno, "Mycorrhizas and nutrient cycling in ecosystems—a journey towards relevance?" *New Phytologist*, vol. 157, no. 3, pp. 475–492, 2003.
- [28] M. J. Swift, O. W. Heal, and J. M. Anderson, *Decomposition in Terrestrial Ecosystems*, Blackwell Scientific, Oxford, UK, 1979.
- [29] J. A. Langley, N. C. Johnson, and G. W. Koch, "Mycorrhizal status influences the rate but not the temperature sensitivity of soil respiration," *Plant and Soil*, vol. 277, pp. 335–344, 2005.
- [30] T. R. Cavagnaro, A. J. Langley, L. E. Jackson, S. M. Smukler, and G. W. Koch, "Growth, nutrition, and soil respiration of a mycorrhiza-defective tomato mutant and its mycorrhizal wild-type progenitor," *Functional Plant Biology*, vol. 35, no. 3, pp. 228–235, 2008.
- [31] J. H. Graham, R. T. Leonard, and J. A. Menge, "Membrane-mediated decrease in root exudation responsible for phosphorus inhibition of vesicular-arbuscular mycorrhiza formation," *Plant Physiology*, vol. 68, pp. 548–552, 1981.
- [32] D. L. Jones, A. Hodge, and Y. Kuzyakov, "Plant and mycorrhizal regulation of rhizodeposition," *New Phytologist*, vol. 163, no. 3, pp. 459–480, 2004.
- [33] M. C. Rillig, S. F. Wright, M. R. Shaw, and C. B. Field, "Artificial climate warming positively affects arbuscular mycorrhizae but decreases soil aggregate water stability in an annual grassland," *Oikos*, vol. 97, no. 1, pp. 52–58, 2002.
- [34] B. Drigo, G. A. Kowalchuk, and J. A. van Veen, "Climate change goes underground: effects of elevated atmospheric CO₂ on microbial community structure and activities in the rhizosphere," *Biology and Fertility of Soils*, vol. 44, no. 5, pp. 667–679, 2008.
- [35] A. H. Fitter, A. Heinemeyer, and P. L. Staddon, "The impact of elevated CO₂ and global climate change on arbuscular mycorrhizas: a mycencentric approach," *New Phytologist*, vol. 147, no. 1, pp. 179–187, 2000.
- [36] I. R. Sanders, R. Streitwolf-Engel, M. G. A. van der Heijden, T. Boller, and A. Wiemken, "Increased allocation to external hyphae of arbuscular mycorrhizal fungi under CO₂ enrichment," *Oecologia*, vol. 117, no. 4, pp. 496–503, 1998.
- [37] J. H. Klironomos, M. F. Allen, M. C. Rillig, et al., "Abrupt rise in atmospheric CO₂ overestimates community response in a model plant-soil system," *Nature*, vol. 433, no. 7026, pp. 621–624, 2005.
- [38] K. K. Treseder, "A meta-analysis of mycorrhizal responses to nitrogen, phosphorus, and atmospheric CO₂ in field studies," *New Phytologist*, vol. 164, no. 2, pp. 347–355, 2004.
- [39] S. Diaz, "Effects of elevated [CO₂] at the community level mediated by root symbionts," *Plant and Soil*, vol. 187, no. 2, pp. 309–320, 1996.
- [40] G. W. T. Wilson, C. W. Rice, M. C. Rillig, A. Springer, and D. C. Hartnett, "Soil aggregation and carbon sequestration are tightly correlated with the abundance of arbuscular mycorrhizal fungi: results from long-term field experiments," *Ecology Letters*, vol. 12, no. 5, pp. 452–461, 2009.
- [41] M. C. Rillig, S. F. Wright, M. F. Allen, and C. B. Field, "Rise in carbon dioxide changes soil structure," *Nature*, vol. 400, no. 6745, p. 628, 1999.
- [42] A. Heinemeyer and A. H. Fitter, "Impact of temperature on the arbuscular mycorrhizal (AM) symbiosis: growth responses of the host plant and its AM fungal partner," *Journal of Experimental Botany*, vol. 55, no. 396, pp. 525–534, 2004.
- [43] C. V. Hawkes, I. P. Hartley, P. Ineson, and A. H. Fitter, "Soil temperature affects carbon allocation within arbuscular mycorrhizal networks and carbon transport from plant to fungus," *Global Change Biology*, vol. 14, no. 5, pp. 1181–1190, 2008.
- [44] C. Plenchette and C. Morel, "External phosphorus requirement of mycorrhizal and non-mycorrhizal barley and soybean plants," *Biology and Fertility of Soils*, vol. 21, no. 4, pp. 303–308, 1996.
- [45] A. Heinemeyer, P. Ineson, N. Ostle, and A. H. Fitter, "Respiration of the external mycelium in the arbuscular mycorrhizal symbiosis shows strong dependence on recent photosynthates and acclimation to temperature," *New Phytologist*, vol. 171, no. 1, pp. 159–170, 2006.
- [46] H. Vierheilig, A. P. Coughlan, U. Wyss, and Y. Piche, "Ink and vinegar, a simple staining technique for arbuscular-mycorrhizal fungi," *Applied and Environmental Microbiology*, vol. 64, no. 12, pp. 5004–5007, 1998.
- [47] S. Declerck, D. G. Strullu, and C. Plenchette, "In vitro mass-production of the arbuscular mycorrhizal fungus, *Glomus versiforme*, associated with Ri T-DNA transformed carrot roots," *Mycological Research*, vol. 100, no. 10, pp. 1237–1242, 1996.
- [48] J. S. King, R. B. Thomas, and B. R. Strain, "Morphology and tissue quality of seedling root systems of *Pinus taeda* and *Pinus ponderosa* as affected by varying CO₂, temperature, and nitrogen," *Plant and Soil*, vol. 195, no. 1, pp. 107–119, 1997.
- [49] E. Kandeler, D. Tschirko, R. D. Bardgett, P. J. Hobbs, C. Kampichler, and T. H. Jones, "The response of soil microorganisms and roots to elevated CO₂ and temperature in a terrestrial model ecosystem," *Plant and Soil*, vol. 202, no. 2, pp. 251–262, 1998.
- [50] I. A. Janssens, M. Crookshanks, G. Taylor, and R. Ceulemans, "Elevated atmospheric CO₂ increases fine root production, respiration, rhizosphere respiration and soil CO₂ efflux in Scots pine seedlings," *Global Change Biology*, vol. 4, no. 8, pp. 871–878, 1998.
- [51] A. Volder, R. M. Gifford, and J. R. Evans, "Effects of elevated atmospheric CO₂, cutting frequency, and differential day/night atmospheric warming on root growth and turnover of *Phalaris* swards," *Global Change Biology*, vol. 13, no. 5, pp. 1040–1052, 2007.
- [52] H. J. De Boeck, C. Lemmens, C. Zavalloni, et al., "Biomass production in experimental grasslands of different species richness during three years of climate warming," *Biogeosciences*, vol. 5, no. 2, pp. 585–594, 2008.
- [53] I. P. Hartley, A. Heinemeyer, and P. Ineson, "Effects of three years of soil warming and shading on the rate of soil respiration: substrate availability and not thermal acclimation mediates observed response," *Global Change Biology*, vol. 13, no. 8, pp. 1761–1770, 2007.
- [54] S. Wan, R. J. Norby, K. S. Pregitzer, J. Ledford, and E. G. O'Neill, "CO₂ enrichment and warming of the atmosphere enhance both productivity and mortality of maple tree fine roots," *New Phytologist*, vol. 162, no. 2, pp. 437–446, 2004.

- [55] M. E. Gavito, P. Schweiger, and I. Jakobsen, "P uptake by arbuscular mycorrhizal hyphae: effect of soil temperature and atmospheric CO₂ enrichment," *Global Change Biology*, vol. 9, no. 1, pp. 106–116, 2003.
- [56] M. Bonkowski, G. Jentschke, and S. Scheu, "Contrasting effects of microbial partners in the rhizosphere: interactions between Norway Spruce seedlings (*Picea abies* Karst.), mycorrhiza (*Paxillus involutus* (Batsch) Fr.) and naked amoebae (protozoa)," *Applied Soil Ecology*, vol. 18, no. 3, pp. 193–204, 2001.
- [57] N. C. Johnson and C. A. Gehring, "Mycorrhizas: symbiotic mediators of rhizosphere and ecosystem processes," in *The Rhizosphere: An Ecological Perspective*, Z. G. Cardon and J. L. Whitbeck, Eds., 2007.
- [58] D. K. Wijesinghe, E. A. John, S. Beurskens, and M. J. Hutchings, "Root system size and precision in nutrient foraging: responses to spatial pattern of nutrient supply in six herbaceous species," *Journal of Ecology*, vol. 89, no. 6, pp. 972–983, 2001.
- [59] P. L. Staddon, A. H. Fitter, and D. Robinson, "Effects of mycorrhizal colonization and elevated atmospheric carbon dioxide on carbon fixation and below-ground carbon partitioning in *Plantago lanceolata*," *Journal of Experimental Botany*, vol. 50, no. 335, pp. 853–860, 1999.
- [60] Y. Luo, S. Wan, D. Hui, and L. L. Wallace, "Acclimatization of soil respiration to warming in a tall grass prairie," *Nature*, vol. 413, no. 6856, pp. 622–625, 2001.
- [61] E. A. Davidson and I. A. Janssens, "Temperature sensitivity of soil carbon decomposition and feedbacks to climate change," *Nature*, vol. 440, no. 7081, pp. 165–173, 2006.
- [62] E. Baath and H. Wallander, "Soil and rhizosphere microorganisms have the same Q₁₀ for respiration in a model system," *Global Change Biology*, vol. 9, no. 12, pp. 1788–1791, 2003.

